

## Data Sheet

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<b>BCRJ Code:</b>	0092
<b>Cell Line:</b>	GK1.5
<b>Species:</b>	Rattus norvegicus (B cell); Mus musculus (myeloma), rat (B cell); mouse (myeloma)
<b>Vulgar Name:</b>	Rat; Mouse, Lewis (B Cell); Balb/C (Myeloma)
<b>Tissue:</b>	Spleen
<b>Cell Type:</b>	Hybridoma: B Lymphocyte
<b>Morphology:</b>	Lymphoblast-Like
<b>Growth Properties:</b>	Suspension
<b>Derivation:</b>	Animals were immunized with the cloned cytotoxic T lymphocyte lines V4 and 243/2.5. Spleen cells were fused with Sp2/O-Ag14 myeloma cells.
<b>Applications:</b>	The antibody profoundly blocks antigen specific murine class II MHC antigen reactive helper T lymphocyte lines.
<b>Products:</b>	immunoglobulin; monoclonal antibody; against mouse helper, inducer T cells (L3T4 antigen, CD4)
<b>Biosafety:</b>	1
<b>Additional Info:</b>	L3T4 is expressed on mouse helper/inducer T cells and is analogous to the human Leu-3/T4 molecule.
<b>Culture Medium:</b>	Iscove's Modified Dulbecco's Medium (IMDM) with 4500 mg/L glucose and fetal bovine serum to a final concentration of 10%.
<b>Subculturing:</b>	Cultures can be maintained by addition of fresh medium. Alternatively, cultures can be established by centrifugation with subsequent resuspension at 1 x 10 <sup>5</sup> viable cells/mL. Maintain cultures at a cell concentration between 1 x 10 <sup>5</sup> and 1 x 10 <sup>6</sup> cells/mL. NOTE: Do not allow the cell concentration to exceed 1 x 10 <sup>6</sup> cells/mL. Population Doubling Time about: 24-30 hours

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**Subculturing****Medium Renewal:**

Every 2 to 3 days

**Culture Conditions:**Atmosphere: air, 95%; carbon dioxide (CO<sub>2</sub>), 5% Temperature: 37°C**Cryopreservation:**

95% FBS + 5% DMSO (Dimethyl sulfoxide)

**Thawing Frozen Cells:**

**SAFETY PRECAUTION:** It is highly recommended that protective gloves and clothing always be used and a full face mask always be worn when handling frozen vials. It is important to note that some vials leak when submerged in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vessel exploding or blowing off its cap with dangerous force creating flying debris. 1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 minutes). 2. Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions. 3. For cells that are sensitive to DMSO it is recommended that the cryoprotective agent be removed immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL complete culture medium and spin at approximately 125 x g for 5 to 7 minutes. 4. Discard the supernatant and resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio). 5. Incubate the culture in an appropriate atmosphere and temperature (see "Culture Conditions" for this cell line). **NOTE:** It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial contents, the culture vessel containing the growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0 to 7.6).

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### References:

Fitch FW, et al. Evidence implicating L3T4 class II MHC antigen reactivity; monoclonal antibody GK1.5 (anti-L3T4a) blocks class II MHC antigen-specific proliferation, release of lymphokines, and binding by cloned murine helper T lymphocyte lines. *J. Immunol.* 131: 2178-2183, 1983. PubMed: 6195255 Dialynas DP, et al. Characterization of the murine T cell surface molecule, designated L3T4, identified by monoclonal antibody GK1.5: similarity of L3T4 to the human Leu-3/T4 molecule. *J. Immunol.* 131: 2445-2451, 1983. PubMed: 6415170 Garvy BA, Harmsen AG. The role of T cells in infection-driven interstitial pneumonia after bone marrow transplantation in mice. *Transplantation* 62: 517-525, 1996. PubMed: 8781619 Murray HW, et al. Models of relapse of experimental visceral leishmaniasis. *J. Infect. Dis.* 173: 1041-1043, 1996. PubMed: 8603949 Wilson ME, et al. Local suppression of IFN-gamma in hepatic granulomas correlates with tissue-specific replication of *Leishmania chagasi*. *J. Immunol.* 156: 2231-2239, 1996. PubMed: 8690913 Wong P, Rudensky AY. Phenotype and function of CD4+ T cells in mice lacking invariant chain. *J. Immunol.* 156: 2133-2142, 1996. PubMed: 8690902 Sayles PC, Johnson LL. Intact immune defenses are required for mice to resist the ts-4 vaccine strain of *Toxoplasma gondii*. *Infect. Immun.* 64: 3088-3092, 1996. PubMed: 8757838 Murray HW, et al. Multiple host defense defects in failure of C57BL/6 ep/ep (Pale Ear) mice to resolve visceral *Leishmania donovani* infection. *Infect. Immun.* 64: 161-166, 1996. PubMed: 8557335 Dialynas DP, et al. Characterization of the murine antigenic determinant, designated L3T4a, recognized by monoclonal antibody GK1.5: expression of L3T4a by functional T cell clones appears to correlate primarily with class II MHC antigen-reactivity. *Immunol. Rev.* 74: 29-56, 1983. PubMed: 6195085 Hay, R. J., Caputo, J. L., and Macy, M. L., Eds. (1992), *ATCC Quality Control Methods for Cell Lines*. 2nd edition, Published by ATCC. Caputo JL. Biosafety procedures in cell culture. *J. Tissue Culture Methods* 11:223-227, 1988 Fleming, D.O., Richardson, J. H., Tulis, J.J. and Vesley, D., (1995) *Laboratory Safety: Principles and Practice*. Second edition, ASM press, Washington, DC. Biosafety in Microbiological and Biomedical Laboratories, 5th ed. HHS. U.S. Department of Health and Human Services, Centers for Disease Control and Prevention. Washington DC: U.S. Government Printing Office; 2007. The entire text is available online at <http://www.cdc.gov/OD/ohs/biosfty/bmbl5/bmbl5toc.htm>

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TIB-207

